

LIMBUS DERIVED STEM CELLS TO TREAT CORNEAL SCARS

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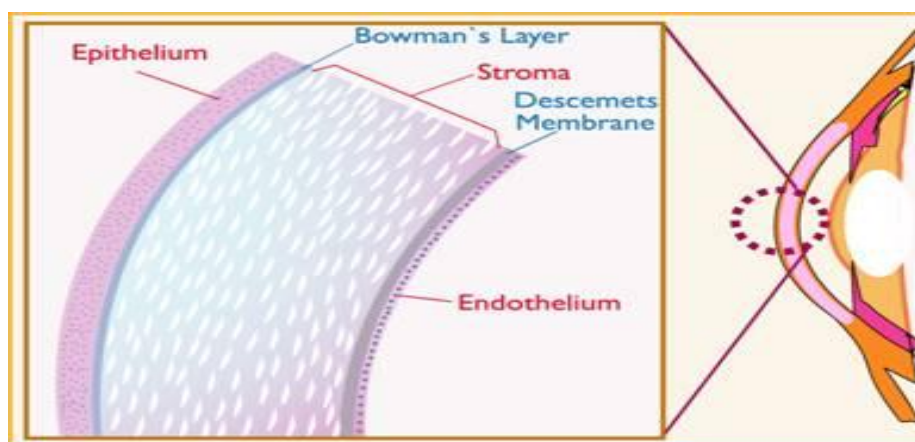
ABSTRACT

Eyes are one of the most precious sensory organs. Eyes are the window of the soul but there are millions of people around the world who are suffering from diseases related to eye. Eyes have an outermost thin layer of tissue known as cornea. The cornea is responsible for 65 – 75 percent of eye's total focusing power, it also protects the eye surface. A corneal dystrophy is a condition in which one or more parts of the cornea lose their normal clarity due to build up of materials that cloud cornea. One of the most common treatments of corneal dystrophy is corneal transplant. A cornea transplant is an operation to remove all or part of a damaged cornea and replace it with healthy donor tissue. Corneas are the most commonly transplanted tissue worldwide. More than 47,000 corneal transplants were being performed in U. S. in 2014. Treatment of corneal dystrophy with corneal transplant suffers from various complications in the patient including tissue rejection, inflammation, shortage of donors etc. A keratoprosthesis (KPro) is an artificial cornea derived from patients own stem cells. A KPro may be the only option available for people who had no success with corneal tissue implants or who have a high risk of tissue rejection. This paper reviews the importance of Limbus derived stem cells as an option for treatment of corneal dystrophy.

KEYWORDS: corneal scars, Limbus, corneal dystrophy, mesenchymal stem cells.**1. INTRODUCTION****1.1 STRUCTURE OF CORNEA**

Cornea is the clear tissue at the front and center of the eye. Its transparency permits light to pass into the eye, through the pupil, lens and onto the retina at the back of the eye. The three major corneal layers are the outer

layer of the cornea or epithelial layer, the middle layer termed the stroma and finally a single layer of cells called the endothelium. The curvature of the cornea plays an important role in focusing light. Its helps protect the eye from infection and foreign material. (Fig.1).

**Fig. 1 Structure of cornea****EPITHELIUM**

The epithelium is the cornea's outermost layer. Its primary functions are to: (a) block the passage into the eye of foreign material, such as dust, water, and bacteria (b) provide a smooth surface to absorb oxygen and nutrients from tears, which are then distributed to the

other layers of the cornea. The epithelium is filled with thousands of tiny nerve endings, that is why our eye may hurt when it is rubbed or scratched. The part of the epithelium that epithelial cells anchor and organize themselves to is called the basement membrane.

BOWMAN'S MEMBRANE

The next layer behind the basement membrane of the epithelium is a transparent film of tissue called Bowman's layer, composed of protein fibers called collagen. If injured, Bowman's layer can form a scar as it heals. If these scars are large and centrally located, they may cause vision loss.

STROMA

Behind Bowman's layer is the stroma, which is the thickest layer of the cornea. It is composed primarily of water and collagen. Collagen gives the cornea its strength, elasticity and form. The unique shape, arrangement and spacing of collagen proteins are essential in producing the cornea's light-conducting transparency.

DESCMET'S MEMBRANE

Behind the stroma is Descemet's membrane, a thin but strong film of tissue that serves as a protective barrier against infection and injuries. Descemet's membrane is composed of collagen fibers that are different from those of the stroma and are made by cells in the endothelial layer of the cornea. Descemet's membrane repairs itself easily after injury.

ENDOTHELIUM

The endothelium is the thin, innermost layer of the cornea. Endothelial cells are important in keeping the cornea clear. Normally, fluid leaks slowly from inside the eye into the stroma. The endothelium's primary task is to pump this excess fluid out of the stroma. Without this pumping action, the stroma would swell with water and become thick and opaque. In a healthy eye, a perfect balance is maintained between the fluid moving into the cornea and the fluid pumping out of the cornea. Unlike the cells in Descemet's membrane, endothelial cells that have been destroyed by disease or trauma are not repaired or replaced by the body.

1.2 CORNEAL DYSTROPHY

A corneal dystrophy is the diseased condition of cornea. These diseases:

- (a) Are usually inherited
- (b) Affect both eyes
- (c) Progress gradually

(d) Don't affect other parts of the body, and aren't related to diseases affecting other parts of the eye or body

(e) Happen in otherwise healthy people.

Corneal dystrophies affect vision in different ways. Some cause severe visual impairment, while a few cause no vision problems and are only discovered during a routine eye exam. Other dystrophies may cause repeated episodes of pain without leading to permanent vision loss. Some of the most common corneal dystrophies include keratoconus, Fuchs' dystrophy, lattice dystrophy, and map-dot-fingerprint dystrophy.

1.2.1 Keratoconus

Keratoconus is a progressive thinning of the cornea. It is the most common corneal dystrophy in the U.S., affecting one in every 2,000 Americans. It is most prevalent in teenagers and adults in their 20s. Keratoconus causes the middle of the cornea to thin, bulge outward, and form a rounded cone shape. This abnormal curvature of the cornea can cause double or blurred vision, nearsightedness, astigmatism and increased sensitivity to light. The causes of keratoconus aren't known, but research indicates it is most likely caused by a combination of genetic susceptibility along with environmental and hormonal influences. About 7 percent of those with the condition have a history of keratoconus in their family. Keratoconus usually affects both eyes. At first, the condition is corrected with glasses or soft contact lenses. As the disease progresses, you may need specially fitted contact lenses to correct the distortion of the cornea and provide better vision. In most cases, the cornea stabilizes after a few years without causing severe vision problems. A small number of people with keratoconus may develop severe corneal scarring or are unable to tolerate a contact lens. For these people, a corneal transplant may become necessary. [Fig 2 (a) & (b)].

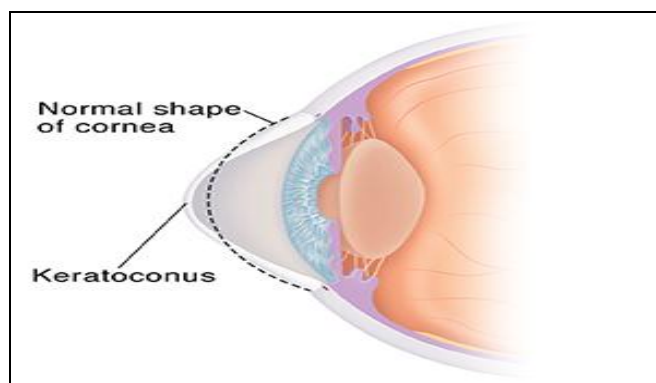


Fig. 2 (a)

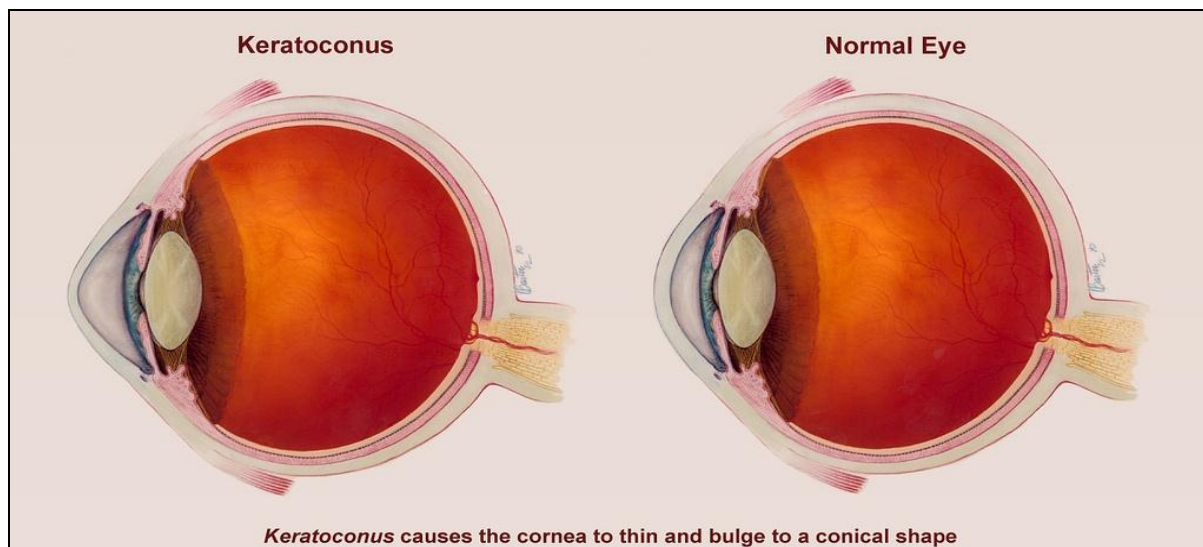


Fig. 2 (a) & 2 (b) Keratoconous cornea

1.2.2 FUCH'S DYSTROPHY

Fuchs' dystrophy is a slowly progressing disease that usually affects both eyes and is slightly more common in women than in men. It can cause vision to gradually worsen over many years, but most people with Fuchs' dystrophy won't notice vision problems until they reach their 50s or 60s. Fuchs' dystrophy is caused by the gradual deterioration of cells in the corneal endothelium; the causes aren't well understood. Normally, these endothelial cells maintain a healthy balance of fluids within the cornea. Healthy endothelial cells prevent the cornea from swelling and keep the cornea clear. In Fuchs' dystrophy, the endothelial cells slowly die off and cause fluid buildup and swelling within the cornea. The

cornea thickens and vision becomes blurred. As the disease progresses, Fuchs' dystrophy symptoms usually affect both eyes and include:

- (a) Glare, which affects vision in low light
- (b) Blurred vision that occurs in the morning after waking and gradually improves during the day
- (c) Distorted vision
- (d) Painful, tiny blisters on the surface of the cornea
- (e) A cloudy or hazy looking cornea (Fig. 3)

The first step in treating Fuchs' dystrophy is to reduce the swelling with drops, ointments, or soft contact lenses. In case of a severe disease, eye care professional may suggest a corneal transplant.

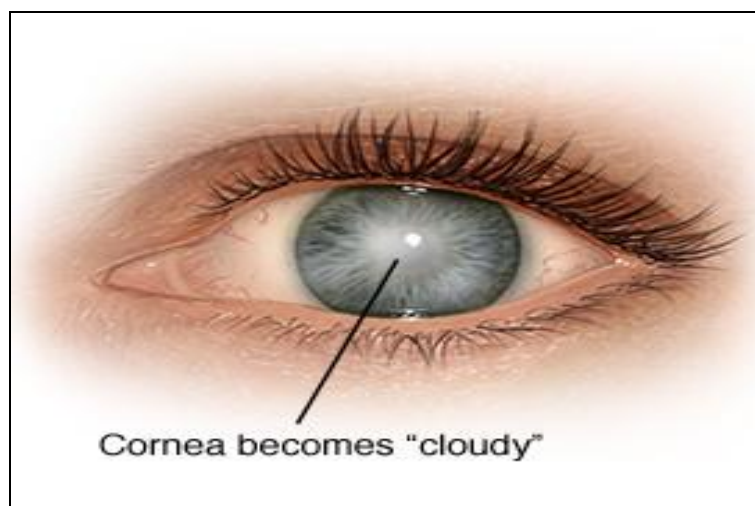


Fig. 3 Fuch's dystrophy

1.3 STEM CELLS TO TREAT CORNEAL SCARS

Treatment of corneal scarring using autologous stem cells, a therapy that, if successful, could reduce the need for corneal grafts. The corneal limbus is the border of the cornea and the sclera (the white of the eye). The limbus is a common site for the occurrence of corneal epithelial

neoplasm. The limbus contains radially-oriented fibrovascular ridges known as the palisades of Vogt that may harbour a stem cell population. The palisades of Vogt are more common in the superior and inferior quadrants around the eye. (Fig. 4) (Fig. 5).

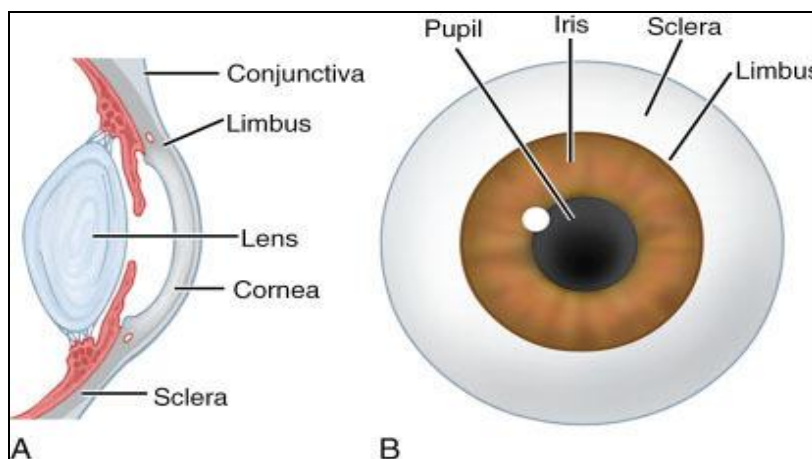


Fig.4 Limbus region of the eye

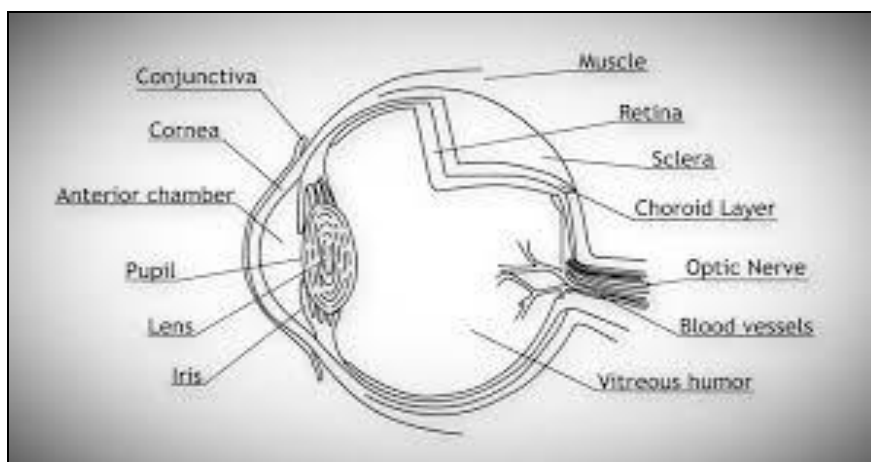


Fig.5 Structure of eye

1.4 TREATMENT OF CORNEAL SCARS

Visual impairment and blindness owing to corneal scarring affect millions of people worldwide (Pascolini *et al.*, & Whitcher *et al.*) and is the commonest indication for corneal transplantation (penetrating keratoplasty) in the developing world. A cornea transplant replaces diseased or scarred corneal tissue with healthy tissue from an organ donor. There are two main types of cornea transplants: traditional, full thickness cornea transplant (also known as penetrating keratoplasty, or PK) and back layer cornea transplant (also known as endothelial keratoplasty, or EK). A graft replaces central corneal

tissue, damaged due to disease or eye injury, with healthy corneal tissue donated from a local eye bank. An unhealthy cornea affects the vision by scattering or distorting light and causing glare and blurred vision. A cornea transplant may be necessary to restore your functional vision. Corneal eye disease is the fourth most common cause of blindness (after cataracts, glaucoma and age-related macular degeneration) and affects more than 10 million people worldwide. Since 1961, more than one million people have had their sight restored with a cornea transplant. (Fig.6).

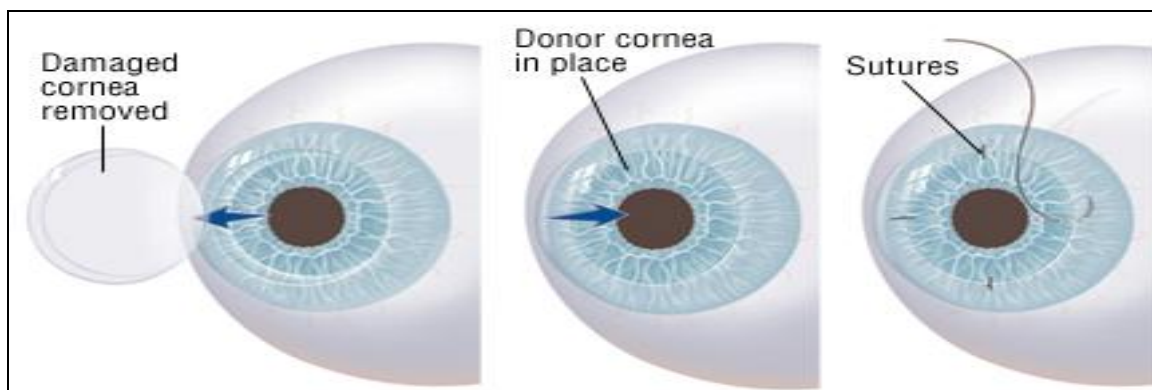


Fig. 6 Penetrating keratoplasty

There have been reports that in a mouse model of corneal opacity, human stromal stem cells were effective in regenerating normal corneal extracellular matrix and repairing collagen fibril defects (Dandona *et al.*). A single population of cells, the corneal stromal stem cells have shown to differentiate to keratocytes and to regenerate stromal tissue *in vivo* (Du *et al.*). Mesenchymal cells obtained from limbal biopsy tissue from human corneas can be expanded in a culture medium containing autologous serum and can differentiate to keratocytes with the potential for use in cell-based therapy.

1.5 CONCLUSION

Over the past decade, characterization of multipotent mesenchymal stem cells in human corneal stroma that differentiate to keratocytes and secrete multilamellar collagenous ECM resembling that of cornea stroma (Hassell *et al.*, Wu *et al.*, (2013), Karamichos *et al.*, Wu *et al.*, (2012) has been done. The studies have revealed two new properties of these cells. First, these cells can be obtained in a biopsy procedure that will permit autologous use. Second, that in an actively healing wound, these cells suppress accumulation of fibrotic scar tissue and promote regeneration of new native, transparent corneal ECM. Corneal stromal stem cells can be obtained from a small surface ocular biopsy. The study have confirmed that mesenchymal stem cells obtained from limbal biopsies are functionally equivalent to corneal stromal stem cells on the basis of sphere-forming ability, gene expression patterns and the ability to organize stromal ECM *in vitro*. In a nutshell, limbus derived stem cells are successful in treating stromal scarring, the major cause of corneal blindness in the world and is a promising treatment for corneal dystrophy.

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